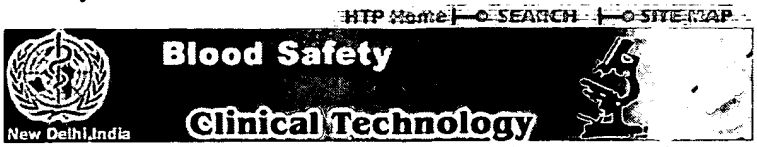


This page must be viewed with IE4 or above, and with Active Scripting enabled.

Sodium and potassium -Flame photometry

•Introduction

Sodium, the major extracellular cation, plays a role in fluid distribution among body compartments. The ingested sodium is filtered in the renal glomerulus and approximately 70% is reabsorbed in the proximal tubule. Further reabsorption occurs in the loop of Henle and <5% is reabsorbed distally under the influence of aldosterone.

About 65-70% of the total body sodium is in its exchangeable form. The exchangeable sodium is made up of extracellular and intracellular sodium. The intracellular sodium concentration is about 10 mmol/L and the extracellular, i.e. the plasma sodium concentration, is about 140 mmol/L. Sodium maintains the osmotic pressure of the extracellular fluid and helps in retaining water in the extracellular compartment. Along with other cations it is also involved in neuromuscular irritability, acid base balance, maintenance of blood viscosity and resting membrane potential.

A high plasma sodium concentration of more than 145 mmol/L is referred to as hypernatremia. This can occur due to simple dehydration, excess sodium intake, steroid therapy as well as in diabetic insipidus. Hyponatremia, with plasma sodium concentration less than 130 mmol/L, can occur due to diuretic medication, kidney disease, excessive sweating, congestive heart failure or gastrointestinal disorder.

Potassium is the major intracellular cation. It is widely distributed in the body in muscle tissue, nerve tissue, blood cells and plasma. It is filtered in the glomerulus, absorbed in the proximal tubule and finally excreted by exchange for sodium in the distal tubule. Potassium influences muscular activity, cardiac function and nerve conduction process.

In hyperkalemia the plasma potassium concentration exceeds 5.5 mmol/L. Acute hyperkalemia is a medical emergency. In hypokalemia the plasma potassium level will be less than 3.5 mmol/L. This can occur due to excessive loss in gastrointestinal secretions and urine, and also in renal tubular acidosis.

•Principle of the method

When a solution of an inorganic salt such as sodium chloride is sprayed into the flame, the elements in the compound are partly converted into the atomic state. Due to the heat energy of the flame a very small proportion of these atoms is excited and the electrons move to a higher energy level. The proportion of the atoms that are excited depends upon the concentration of the particular element and on the temperature of the flame. In the excited state the electrons are unstable and they rapidly revert back to their former lower energy level. As they change from the excited state or higher energy level back to the lower energy level, they emit the light in the form of a fixed wavelength, to produce a spectrum. Under carefully controlled conditions the amount of light emitted is directly proportional to the number of atoms that are excited, which in turn is proportional to the concentration of the substance in the sample.

•Specimen type, collection and storage

Both sodium and potassium are stable in serum for several hours at 25-35° C and for 3 months at -20° C. Anticoagulants containing sodium or potassium salts are not suitable, but lithium heparin may be used as an anticoagulant.

If whole blood is left unseparated for >3hours or refrigerated, potassium will leak out of the red cells giving falsely increased values.

•Reagents

All chemicals must be Analar grade.

Sodium chloride (NaCl) and potassium chloride (KCl) should be dried for 2-3 hours at about 100° C before use. Before weighing, the chemicals must be allowed to cool to room temperature either in a desiccator or in a container with a tight-fitting lid with a small air space.

▶ Stock Sodium 1000 mmol/L

Weigh out 29.25 g dried NaCl, dissolve in about 400 ml of distilled water taken in a 500 ml volumetric flask and then make up to 500ml with distilled water. Store in a pyrex glass bottle at 25-35° C. Stable for one year.

▶ Stock Potassium 100 mmol/L

Weigh out 0.746 g dried KCl, dissolve in about 80 ml of distilled water taken in a 100 ml volumetric flask and then make up to 100 ml with distilled water. Store in a pyrex glass bottle at 25-35^{0}C. Stable for one year.

▶ Working Standards

- Low standard for Sodium 100mmol/L: Dilute 10 ml of stock sodium to 100 ml with distilled water. Stable for 6 months at 25-35^{0}C.
- Combined standard for sodium and potassium 140Na^+/5K^+ mmol/L: Dilute 14 ml of stock sodium and 5 ml of stock potassium together to 100 ml with distilled water. Store in a pyrex bottle at 25-35^{0}C. Stable for 6 months.
- Aspiration standard for sodium - 1.0 mmol/L: Dilute 1.0 ml of working standard to 100 ml with distilled water. Prepare fresh each time.
- Combined aspiration standard for sodium 1.4 mmol/L and potassium 0.05 mmol/L. Dilute 1.0 ml of working standard (combined standard for Na^+/K^+140/5mmol/L) to 100ml with distilled water. Prepare fresh each time.

•Equipment, glassware and other accessories

Refer to Section A (2), Introduction to SOP.

•Procedure

The protocol of the procedure is described below.

▶ Sample dilution

Dilute each serum sample 1:100 with distilled water by mixing 0. 1ml sample with 9.9 ml distilled water.

▶ Procedure for simultaneous measurement of Na+ & K+ in the flame photometer (digital flame photometer)

- Switch on the flame photometer. Digital display should turn on.
- Turn the set '(full scale) F.S. coarse and fine controls' into maximum clockwise position.
- Select appropriate filter with the help of filter selector wheel (Na^+ on the left side and K^+ on the right side).
- Switch on the compressor and check the air pressure. Adjust it to read between 0.4 and 0.6 k g/cm^2.
- Open the gas cylinder, remove the trapper at the rear of the flame photometer and ignite the flame.
- Adjust the gas regulator to get a maximum height non-luminous blue flame with 10 distinct cones (5 on each side of the burner head).
- Feed distilled water to the atomizer and wait for at least 30 seconds.
- Adjust the 'Set Ref Coarse' and Fine controls' to zero digital readout for K^+ only.
- Aspirate 1.0 mmol/L Na^+ solution. Wait at least 30 seconds and then adjust the Set Ref Coarse and Fine controls' to a digital read out of I 00 for Na^+ only.
- Aspirate the combined standard solution (1.4/0.05, Na^+/K^+) and wait at least for 30 seconds. Adjust 'F.S control' on Na^+ side for readout 140 and that on K^+ side for a digital readout of 50.
- Repeat steps 9 and 10 once again. The flame photometer now stands calibrated.
- Now feed diluted test sample / QC to the atomizer for at least 30 seconds before recording the readings for Na^+ and K^+.

• Calculation

After aspirating the standard solution, the digital reading for Na^+ is adjusted to 140 and that of K^+ to 50. This is done in order to represent Na^+ and K^+ values in undiluted serum. Since the test sample/QC is diluted initially 1: 100 and then aspirated, the initial standard values for Na^+ & K^+ (1.4 & 0.05 mmol/L) must be multiplied by 100 to represent 140 mmol/L Na^+ and 5 mmol/L K^+. In the case of K^+, in order to improve the sensitivity of the assay the digital reading for the standard is further multiplied by 10 to show a reading of 50.

In essence, the test sample/QC digital readings are compared with the standard readings for Na^+ and K^+. The digital reading appearing for Na' of the test sample/QC is read as mmol/L value straightaway. On the other hand, the test sample /QC K^+ value represents 1/10 th of the digital reading.

For example, digital reading for "140 Na^+ = 140 mmol/L Na^+; digital reading for 45 K^+ = 4.5 mmol/L K^+

• Analytical reliabilities

Refer to **pages 7-9 of section 1 (General Introduction)** on the use of internal QC and interpretation of daily QC data (for releasing patients' results).

Since Na^+, K^+ are very commonly analysed parameters in a laboratory, it is recommended that internal

QC (normal QC pool) be included with every batch of samples analysed in a day, irrespective of the number of samples in a batch. Further, even when a single sample is analysed as an "emergency" sample at any time of the day or night, it is essential to include an internal QC. From the QC results obtained for the day, mean, standard deviation and % CV can be calculated to ensure that ***within-day precision*** is well within the acceptable limit, i.e. 4%.

The mean value of internal QC for the day can be pooled with the preceding 10 or 20 mean values obtained in the previous days and ***between-day precision*** can be calculated and expressed as % CV. Ensure that this is well within the acceptable limit, i.e, 8%.

At least once a week analyse another QC serum from either a low QC or high QC pool.

"Assayed" QC sera with stated values (ranges) are available from several commercial sources, viz. Boehringer Mannheim, BioRad & Randox.

However, care should be taken when operating the flame photometer as the technician will be using liquefied petroleum gas. Leakage of either air or gas during operation will cause explosion. Apply soap solution at the connecting point to check such leakage.

•Hazardous materials

The reagents used are made up of only sodium chloride and potassium chloride. Therefore no precautionary measures are required.

•Reference range and clinical interpretation

The reference ranges by this method are:

- Serum sodium 130 - 145 mmol/L
- Serum potassium 3.5 - 5.0 mmol/L

Elevated levels of serum sodium occur in conditions such as severe dehydration, hyperadrenalism and brain injury.

Low serum sodium values are noticed in metabolic acidosis, salt- losing nephritis, Addison's disease, etc. Increased serum potassium level is observed in anoxia, metabolic renal tubular acidosis and shock or circulatory failure.

Low serum potassium values are observed due to low intake of dietary potassium over a period of time or increased loss through kidney, vomiting or diarrhoea. Increased secretion of adrenal steroids or some diuretics may also promote the loss of potassium.

•Limitations

Avoid using haemolysed serum. This will cause elevated K^+ level.

Reliability of the results depends on the proper maintenance of the flame photometer, salient features of which are listed below.

- **Requirements**

•Non-luminous blue flame
•Supply of dry air at a controlled pressure, viz. 10- 15 Kg /cm2
•Regular availability of liquid petroleum gas

▶ Maintenance

- Disconnect power and gas supply before proceeding to do maintenance.
- Turn the control in both Na+/K+ display fully anti-clockwise.
- Disconnect the drain outlet and the gas and air inlets.
- Remove the top panel and disconnect the photocell.
- Remove the side panel and take out the atomizer.
- Disconnect the air line at the pressure gauge.
- Disconnect the air and gas inlets to the mixing chamber.
- Remove the burner head and remove mixing chamber
- Wash the above well with tap water and distilled water.
- Clean the atomizer with a thin wire and adjust its spray by passing compressed air. This can be done by screwing / unscrewing the two knurled nuts in the atomizer.
- Remove the air tube and flush out any water remaining in the tube due to the cooling of compressed air.
- Do not use oxyacetylene or highly explosive mixture as fuel.
- After cleaning, refix everything carefully.

▶ Cleaning of various units in a flame photometer

- ***Atomizer and capillary tube*:** Flushing with copious amount of distilled water is adequate. If blockage occurs, remove the atomizer from its seating and flush with dry air or clean it using a thin wire. If cleaning of atomizer is done with a wire before and after using it, blockage will rarely occur. If all the above fails, a new atomizer is to be fixed.
- ***Mixing chamber:*** Flushing with distilled water is adequate. Do not use detergent or soap solution because it will remain inside if washing is not done properly out and will give erratic reading due to the presence of Na+ / K+ in the soap solution.

▶ Fault diagnosis

Symptom	Diagnosis	Remedy
1. Unstable reading	Excessive vibration	Provide shock-proof base (e.g.) glass plate on foam rubber.
	Air supply blocked	Check air supply and clear blockage
	Atomizer low gas pressure	Remove, wash and dry burner
	Filter dirty	Clean with isopropanol.
2. Intermittent reading	Blocked atomizer	Clean atomizer using a thin wire
	Faulty photocell	Change photocell
	Dirty photocell	Clean photocell
3. Low sensitivity	Blocked atomizer	Clean blockage
	Low gas pressure	Check gas pressure
	Faulty photocell	Change photocell.

| Foreword | Acknowledgements | Preface | General Introduction | Biochemical Measurements in Plasma/Serum | Biochemical Measurements in CSF | Urinalysis |
| WHO/SEARO Home | SEARO Search | SEARO Sitemap |Contact Us |

Interdisciplinary Modules to Teach Waste or Residue Management in the Food Chain

MODULE 2: IDENTIFICATION, QUANTIFICATION, AND CHARACTERIZATION OF WASTES/RESIDUES

Text Only Module 2

CHARACTERIZATION

Physical Characteristics | Chemical Characteristics | Microbiological Characteristics

Introduction
General Characterization
Identification
Quantification
Characterization
References
Home
Module Description
Instructor's Manual

Chemical Characteristics

Multiple methods for measuring the chemical content of food products have been approved by AOAC. The AOAC method specific to the food product being tested should be selected. A few specific examples are included in the following section:

- Moisture Content
- Proximate Analysis
- Nutritional Analysis
- Energy

Moisture Content

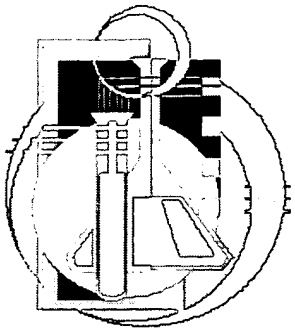
Wet-weight method of measurement. Moisture is expressed as a percentage of the wet weight of the material. It is used most commonly in the field of solid waste management (Theodore & Theodore, 1996). See Appendix 2D-2 for the formula.

- AACC Method 44-15A (Moisture – Air-Oven Methods) (AACC, 2000) for moisture content (wet basis) of the product samples.
- AOAC Method no. 925.10 (Air Oven Method) for moisture in flour (AOAC, 2000).
- AOAC Method no. 985.14 (Rapid Microwave Drying Method) for moisture in meat and poultry products (AOAC, 2000).
- AOAC Method no. 920.36 for moisture in animal feed (AOAC, 2000).

Dry-weight method of measurement. Moisture is expressed as a percentage of the dry weight of the material (Theodore & Theodore, 1996). See Appendix 2D-2 for the formula.

Back to the top...

Proximate Analysis



Proximate analyses include the determination of protein, crude fat, fiber, and mineral contents. The carbohydrate content is usually determined by the difference from the original sample minus the moisture, protein, crude fat, and mineral content, with their contents calculated at the same moisture level. The following methods are indicated as a reference. Professional organizations have developed their own repeatable and reliable methods. In waste/residue characterization studies, it is important to use the same method when comparing samples.

Protein content.

- AOAC Method no. 992.15 (Combustion Method-Leco FP428) for crude protein in meat and meat products (AOAC, 2000).
- AOAC Method no. 950.36 for protein in baked products (AOAC, 2000).
- AOAC Method no. 990.03 (Combustion Method) for protein in animal feed (AOAC, 2000).
- AOAC Method no. 984.13 (Copper catalyst Kjedahl method) for protein in animal feed (AOAC, 2000).

Crude fat content.

- AACC 30-25 (Crude fat in wheat, corn, and soy flour, feeds, and mixed feeds) (AACC, 2000).
- AOAC Method no. 991.36 (Solvent Extraction Method) for fat in meat (AOAC, 2000).
- AOAC Method no. 960.26 (Rapid Detergent Method) for fat in raw milk (AOAC, 2000).
- AOAC Method no. 920.39 for crude fat in animal feed (AOAC, 2000).

Fiber content.

- AOAC Method no. 920.86 for crude fiber in flour (AOAC, 2000).
- AOAC Method no. 950.37 for fiber (crude) in bread (AOAC, 2000).
- AOAC Method no. 991.43 (Enzymatic-Gravimetric method – TRIS Buffer) for total, soluble, and insoluble dietary fiber in foods (AOAC, 2000).
- AOAC Method no. 962.09 (Ceramic fiber filter method) for crude fiber in animal feed (AOAC, 2000).
- The Van Soest detergent system is used to determine Neutral Detergent Fiber (NDF), Acid Detergent Fiber (ADF), and lignin fractions of the masa coproduct samples. These methods are widely used for the determination of the fiber composition of biological materials and potential livestock feed ingredients (Rosentrater et al., 1999).

Carbohydrate content.

- AOAC Method no. 971.18 (Gas Chromatographic Method) for carbohydrates in fruit juices (AOAC, 2000).
- The carbohydrate content is often determined as the difference between the protein, moisture, fat, and ash percentages (Ferris, et al, 1995). The sum of the

percentages of protein, moisture, fat, and ash is subtracted from 100 to determine the percentage of carbohydrate in the sample.

Back to the top...

Nutritional Analysis

Amino acid composition.

- AOAC Method no. 999.13 (Amino Acid Analyzer) for Lysine, Methionine, and Threonine in Feed Grade Amino Acids and Premixes (AOAC, 2000).

Calcium content.

- AOAC Method no. 991.25 (Atomic Absorption Spectrophotometric and Colorimetric Method) for calcium in cheese (AOAC, 2000).
- AOAC Method no. 935.13 for calcium in animal feed (AOAC, 2000).
- Calcium and magnesium: APHA Method Standard Methods for the Examination of Water and Wastewater (SMEWW) 3111 B, which utilizes flame atomic absorption spectrophotometry, and potassium analysis conducted using Method SMEWW 3500-K D, which utilizes flame photometry (APHA, 1995).

Magnesium content.

- AOAC Method no. 931.10 for magnesium in fruits and fruit products (AOAC, 2000).
- AOAC Method 2.6.01 and AOAC Method 968.08 (Atomic Absorption Spectrophophosphometric Method) for magnesium in Animal feed (AOAC, 2000).

Phosphorus content.

- AOAC Method no. 991.27 (Spectrophotometric Method) for phosphorous in meat and meat products (AOAC, 2000).
- AOAC Method no. 931.06 for phosphorous in Eggs (AOAC, 2000).
- AOAC Method 965.17 (Photometric method) for phosphorous in animal feed (AOAC, 2000).

Potassium content.

- AOAC Method (Flame emission spectrometric method) no. 990.23 for sodium and potassium in dried milk (AOAC, 2000).
- AOAC Method (Chloroplatinate method) no. 929.05 for potassium in fruits and fruit products (AOAC, 2000).
- AOAC Method no. 969.04 for potassium in fertilizers (AOAC, 2000).

Back to the top...

Energy

The energy content of a sample can be estimated if the percentage of protein, fat, and carbohydrates is known. The conversion factors used are 16.74 kJ (4.0 kcal)/g for protein and carbohydrates and 37.66 kJ (9.0 kcal)/g for fat (Whitney, et al, 1987; Ferris et al., 1995). A bomb calorimeter can be used to measure the total energy for a food product.

Some of figures, tables, and appendixes are in PDF format. If you do not have a PDF reader, you can download the Adobe version from the Acrobat Download Page.

Back to the top...

Introduction | General Characterization | Identification | Quantification | Characterization | References

Home | Module Description | Module 1 | Module 2 | Module 3 | Module 4 | Instructor's Manual

APPENDIX 2D

Formula for Characterization of Wastes/ Residues

2D-1. Particle size. The size of the waste component can be computed using length, height, or width. The following formula can be used to calculate size of component (Theodore & Theodore, 1996).

$$Sc = l$$

$$Sc = \frac{l + w}{2} l$$

$$Sc = \frac{l + w + h}{3} l$$

where
Sc = size of component, in (mm)
l = length, in (mm)
w = width, in (mm)
h = height, in (mm)

The ASAE standard 319.3 JUL 97 (ASAE, 2000) recommends that the size of dry particulates be reported as geometric mean diameter and geometric standard deviation by weight. The following formula can be used to compute these values.

$$d_{gw} = \log^{-1}\left[\frac{\sum_{i=1}^{n}\left(W_i \log \overline{d_i}\right)}{\sum_{i=1}^{n} W_i}\right]$$

$$S_{\log} = \left[\frac{\sum_{i=1}^{n} W_i\left(\log \overline{d_i} - \log d_{gw}\right)^2}{\sum_{i=1}^{n} W_i}\right]^{1/2} = \frac{S_{\ln}}{2.3}$$

$$S_{gw} \approx \frac{1}{2} d_{gw}\left[\log^{-1} S_{\log} - (\log^{-1} S_{\log})^{-1}\right]$$

where :

d_i is nominal sieve aperture size of the i^{th} sieve, mm

d_{i+1} is nominal sieve aperture size in next larger than i^{th} sieve (just above in a set), mm

d_{gw} is geometric mean diameter or median size of particles by mass, mm
is geometric diameter or median size of particles on sieve, mm
is $(d_i \times d_{i+1})^{1/2}$

S_{log} is geometric standard deviation of log-normal distribution by mass in ten-based logarithm, dimensionless

S_{ln} is geometric standard deviation of log-normal distribution by mass in natural logarithm, dimensionless

S_{gw} is geometric standard deviation of particle diameter by mass, mm

W_i is mass on i^{th} sieve, g

n is number of sieves + 1 (pan)

2D-2. Moisture Content. The moisture content calculation is done using the following equation.

1) Wet weight method of measurement

$$M = \frac{w - d}{d} \times 100$$

where
M = moisture content, %
w = initial weight of sample as delivered, lb (kg)
d = weight of sample after drying at 105 °C, lb (kg)

2) Dry weight method of measurement

$$M = \frac{w - d}{w} \times 100$$

where
M = moisture content, %
w = initial weight of sample as delivered, lb (kg)
d = weight of sample after drying at 105 °C, lb (kg)